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Thermoplastic extrusion technology as a tool for adding value to brewer's by-products

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ABSTRACT

Brewer's by-products show high nutritious potential to be reintegrated into the food chain. Nevertheless, their high moisture, microbiological instability, and heterogeneity pose enormous challenges. This study investigated thermoplastic extrusion to repurpose large amounts of the two main brewery by-products: brewer's spent grains (BSG) and brewer's spent yeast (BSY). A mixture design was used to compose ten blends with different proportions of dried-BSG, BSY, and corn grits, maximizing the use of BSG up to 70 g/100 g. Extruder parameters were fixed. PLS regression was applied to evaluate the effects of blend composition on the characteristics of the extruded products. Thermoplastic extrusion shows feasibility in transforming large volumes of by-products into value-added products. Blends with high insoluble fiber, protein, and moisture contents resulted in extrudates with high protein concentration, low expansion, and high density. The results of this exploratory work can be used for product optimization by choosing nutritional, technological, and commercial targets to create innovative, healthy, and sustainable food products that meet consumer demands.

1. Introduction

In times when attempts are made to manage the generation and disposal of domestic and industrial waste to alleviate the environmental impact of human activities, repurposing side-streams from the food industry is an important alternative (Sahin et al., 2021). Various nutrients could be introduced or reinserted into the food chain through specific processes, increasing the food supply using low-cost raw materials of high nutritional value (REPRO, 2008). Moreover, developing new products with agro-industrial by-products meets consumer trends toward more sustainable and healthier products (Euromonitor, 2018).

Brewer's spent grains (BSG), usually sold as animal feed, accounts for 85% of the brewery's by-products (Mussato et al., 2006). Considering an average 23 kg of BSG per beer hectoliter (BA, 2016), we estimate a world production of 42.8 million tons of BSG in 2021 (Barth, 2023).

BSG is mainly composed of malt husks, containing about 70 g/100 g fibers and 30 g/100 g proteins, on a dry basis, besides minerals, vitamins, phenolic acids, and essential fatty acids. Its composition depends mostly on beer formulation, processing characteristics, and handling of by-products. However, it contains 70–80 g/100 g of moisture and, due to its residual starch/sugar and peptide content, it is highly susceptible to spoilage (Ikram et al., 2017; Lynch et al., 2016; Mussato et al., 2006).

BSG's great potential lies in its composition of fibers, which includes cellulose, hemicellulose, lignin, arabinoxylans, and beta-glucan, which are related to health benefits. Moreover, its amino acid composition comprehends 30 g/100 g of essential amino acids, with a remarkable lysine content, different from most cereal-based materials (Sahin et al., 2021 ab).

Most studies aiming at BSG's valorization focus on the extraction of its most valuable compounds or BSG biotechnological utilization, although the incorporation of up to 30 g/100 g dried BSG into bread, cookies, pasta, and ready-to-eat snacks has been well explored (Sahin et al., 2021a,b, Puligundla; Mok, 2021; Bonifácio-Lopes et al., 2020; Ikram et al., 2017; Nascimento et al., 2017; Lynch et al., 2016; Stojceska et al., 2008; 2009; Mussato et al., 2006). Reis and Abu-Ghannam (2014) reported an increase in phenolic content and antioxidant capacity, and a decrease in glycemic index with an addition of 40 g/100 g BSG to extrudates.

Brewer's spent yeast (BSY) is the second most generated by-product in the brewing industry, estimated at 1.5–1.8 kg BSY per beer hectoliter, summing about 2.8–3.3 million tons of BSY in 2021. This by-product, with a dry matter of 10–14 g/100 g, including yeast and beer solids,

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has exceptionally high chemical oxygen demand, causing huge problems if mixed in wastewater (Ferreira et al., 2010; Jaeger et al., 2020).

BSY is a valuable by-product since it is an excellent source of proteins, flavor enhancers, B-complex vitamins, nucleic acids, minerals, β-glucans, and mannoprotein. The type of fermentation, besides the yeast strain, beer style, and whether the yeast is vital or exhausted, determine BSY composition (Charlton & Vriesekoop, 2017; Jaeger et al., 2020; Ferreira et al., 2010).

Yeast extract is widely applied as a flavor enhancer and food supplement. However, the level of nucleic acids, primarily RNA, limits BSY use as a protein supplement due to its detrimental effects on uric acid. Despite the several potential uses for BSY, its primary use is as a cheap source of protein for animal feed (Ferreira et al., 2010; Jaeger et al., 2020).

Besides the high risk of spoilage, the high moisture levels of these by-products make storage and transportation costly (Lynch et al., 2016). The most studied techniques for valorizing by-products include extracting isolated compounds such as micro and macronutrients, gelling agents, hydrocolloids, fiber fractions, and bioactive compounds. Despite generating high added-value outputs, these processes do not utilize the whole by-product, only reducing waste. Consequently, there is still a demand for economically viable systems that use large volumes of by-products.

Thermoplastic extrusion is a technique that combines high shear rates, temperature, and pressure to cause transformations in the raw material, resulting in products with different shapes and textures, depending on the process conditions and composition of the ingredients. It is extensively applied to produce ready-to-eat snacks, breakfast cereals, food ingredients, meat analogs, and animal feed. The extrusion process allows the usage of materials with different compositions, rich in starch, proteins, or fibres, each one generating products with unique characteristics. It can be used for large production volumes, has high productivity, is economical, and does not generate new waste, presenting enormous potential in agro-industrial by-product reuse (Steel et al., 2012).

Both the composition of the raw material and the process parameters directly influence the properties of the extrudate. It is necessary to carefully choose the feed moisture, the chemical composition of the ingredients (amount and type of starch, proteins, fats, and sugars), its physical state and particle size, feed rate, feeder configuration, the temperature of each extruder module, speed and screw configuration, the length of the extruder and the geometry of the die. All these parameters will determine the extent of the modifications that the material will undergo during the process, influencing the rheological properties of the fluid melt inside the extruder and, consequently, the characteristics of the extruded product (Fellows, 2000; Meng et al., 2010).

Considering the amounts of by-products generated yearly, it is crucial to propose and study processes and techniques that upcycle large proportions of them. This study explores thermoplastic extrusion to repurpose the two main brewery by-products, contributing to waste reduction, the sustainability of the brewing industry, and offering insights to develop innovative, healthier, and more sustainable foods with low-cost ingredients.

We prepared blends with high concentrations of brewer's spent grains (BSG) in combination with brewer's spent yeast (BSY), and corn grits, maximizing the first and minimizing the latter to keep moisture below the processing limit of 30 %. We aimed to investigate the feasibility of working with up to 80 g/100 g of brewer's by-products and report the effects of blend composition on the extruded products' technological attributes, besides the challenges and solutions for working with by-products with high fibre, protein, and moisture content.

The findings of this exploratory work provide insights for future product optimization, targeting nutritional, technological, and commercial objectives in the pursuit of a desirable product.

2. Material and methods

2.1. Raw material

Brewer's spent grains (BSG) from a *Pilsen-type* filter-press beer process, with a composition of 18.25 g/100 g proteins, 9.84 g/100 g digestible carbohydrates, 54.52 g/100 g total dietary fibers, 13.58 g/100 g lipids, 3.81 g/100 g ash, in dry basis, and a moisture content of 78.57 g/100 g; brewer's spent yeast (BSY), composed of 53.25 g/100 g proteins, 17.55 g/100 g digestible carbohydrates, 19.88 g/100 g total dietary fibers, 2.52 g/100 g lipids, 6.79 g/100 g ash, dry basis, with a moisture content of 86.46 g/100 g; and corn grits (GRITS), containing 7.34 g/100 g proteins, 89.66 g/100 g digestible carbohydrates, 2.63 g/100 g lipids, 0.36 g/100 g ash, in dry basis, and a moisture content of 11.34 g/100 g, were donated by a Brazilian brewery. The chemical composition of the raw material was assessed in our previous work (Brito et al., 2018).

To maximize BSG usage and avoid spoilage, it was dried at 90 °C for 12h, to a moisture content of 10 g/100 g, before extrusion.

2.2. Mixture design

A mixture experiment was applied to evaluate how different proportions of dried BSG (x_1), BSY (x_2), and GRITS (x_3) would affect the properties of the extruded products. A modified simplex centroid design with three components (Fig. 1) was selected, totalizing twelve experiments (including three replicates of the centroid point to evaluate process reproducibility). Each mixture component is expressed by a fraction, and their sum is a unity ($x_1 + x_2 + x_3 = 1$) (Analytics, 2017; Cornell, 2011, pp. 1–93; Eriksson et al., 1998).

Based on pre-tests, lower limits (Table 1) were established for each component of the mixture to:

(i) ensure that all three components were present in each blend (Cornell, 2011, pp. 1–93);
(ii) maximize the use of BSG - the by-product generated in the highest amounts during the brewing process;
(iii) minimize the use of BSY - the most valuable of the by-product and the one with the highest moisture content;
(iv) minimize the use of GRITS - starchy material with low moisture content added to improve the extrusion process and the technological properties of the extrudates;

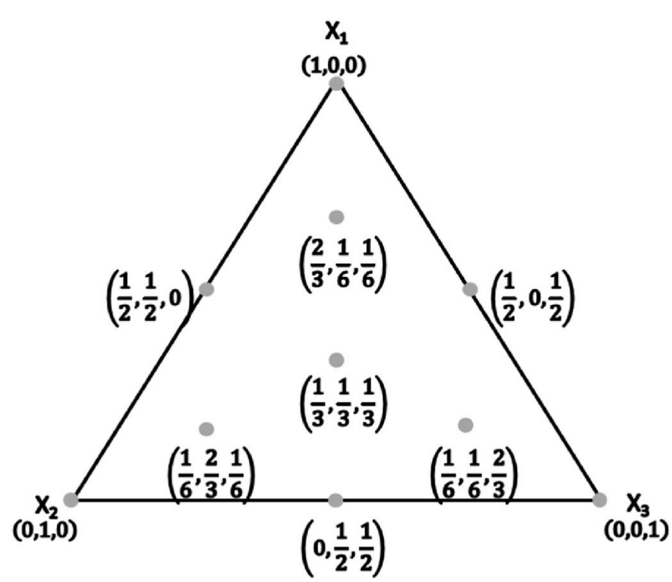

Fig. 1. Modified simplex-centroid with 3 components.

Table 1
Lower limits of the mixture components.

	Component	Lower Limits
DBSG	x_1	0.4
BSY	x_2	0.1
GRITS	x_3	0.2

*BSG = dried brewer's spent grains; BSY = brewer's spent yeast; GRITS = corn grits.

(v) do not exceed 30 g/100 g of moisture in the blends - maximum limit for a stable and homogeneous feed flow in the extruder.

The original simplex design with three components is represented by a triangle (Fig. 1), where in each vertex the mixture is composed entirely by one component (e.g., $x_1 = 1$, $x_2 = 0$, $x_3 = 0$). The use of lower limits implies working with a subregion of the triangle. To facilitate understanding the simplex and fitting of the model, L-pseudocomponents are employed, representing the new limits (Table 2). Eq. (1) is applied to calculate the real concentrations, based on the L-pseudocomponents (Cornell, 2011, pp. 1–93):

$$x_i = L_i + (1 - L)x_i^{'} \tag{1}$$

Where, x_i is the design original component, L_i is the lower limit and L is the sum of the lower limits of the variables (L = 0.4 + 0.1 + 0.2 = 0.7), $x_i^{'}$ is the L-pseudocomponent, resulting in Eq. (2) a, b, c.

$$\text{a)}x_1 = 0,4 + x_1^{'}(1 - 0,7)\text{b)}x_2 = 0,1 + x_2^{'}(1 - 0,7)\text{c)}x_3 = 0,2 + x_3^{'}(1 - 0,7) \tag{2}$$

Table 2 shows the L-pseudocomponents and the real concentrations of each variable. We used the real concentrations to calculate the effect of the blends' composition on the blends' moisture content. Only Blend 2 would exceed the limit of 30 %, reaching a calculated moisture content of 40.85 %. The other blends were expected to have 17.9 %–29.58 % moisture, including the centroids of the simplex with 25.69 %.

2.3. Thermoplastic extrusion process

The extrusion process was conducted in a ZSK 30 twin-screw extruder (Werner & Pfleiderer Corp., Ramsey, USA), with a 29 L/D (length/diameter) ratio, coupled to a K-Tron k2 volumetric feeder (K-Tron North America, Pitman, NJ, USA). To evaluate the effect of the blend composition the process conditions were fixed: feed screw speed 250 rpm; screw speed 390 rpm; temperature of the last zone of the extruder 110 °C; die diameter 4.8 mm. The screw configuration was composed by forward and neutral elements. After extrusion, the products were oven-dried at 80 °C for 30 min. Extrusion conditions were chosen based on pre-tests with the experiments corresponding to the centroid of the simplex.

Table 2
L-pseudocomponents and real concentrations of the mixture design with 3 components: dried brewer's spent grains (BSG), brewer's spent yeast (BSY) and corn grits (GRITS), with pre-stablished lower limits.

	L-pseudocomponents			Real concentrations		
Experiment	x'1	x'2	x'3	BSG (x_1)	BSY (x_2)	GRITS (x_3)
1	1	0	0	0.70	0.10	0.20
2	0	1	0	0.40	0.40	0.20
3	0	0	1	0.40	0.10	0.50
4	1/2	0	1/2	0.55	0.10	0.35
5	0	1/2	1/2	0.40	0.25	0.35
6	1/2	1/2	0	0.55	0.25	0.20
7	2/3	1/6	1/6	0.60	0.15	0.25
8	1/6	2/3	1/6	0.45	0.30	0.25
9	1/6	1/6	2/3	0.45	0.15	0.40
10	1/3	1/3	1/3	0.50	0.20	0.30
11	1/3	1/3	1/3	0.50	0.20	0.30
12	1/3	1/3	1/3	0.50	0.20	0.30

x'1, x'2, x'3 = BSG, BSY and GRITS L-pseudocomponents, respectively. x1, x2, ×3 correspond to their real values. DSG = dried spent grain; BSY = brewer's spent yeast; GRITS = corn grits. L-pseudocomponents and real concentrations for each experiment sum a unit. The former is expressed as fractions, while the latter in decimal form.

2.3.1. Feed Rate (FR)

The feed rate (FR – kg/h) was measured for each blend at different feed-screw speeds. Linear equations were used to estimate linearity and calculate FR at process feed speed.

2.3.2. Specific Mechanical Energy (SME)

SME was calculated for each experiment through Eq. (3) (Stojceska et al., 2008):

$$SME(kW.h/kg) = \frac{\text{screw speed (rpm)xmotor power (kW)xtorque (\%)}}{\text{maximalscrew speed (rpm)xfeed rate (kg/h)x 100}} \tag{3}$$

where, screw speed: 397 rpm; maximum motor power: 9.1 kW; maximal screw speed: 500 rpm; torque: provided by the system; feed rate: calculated (item 2.4).

2.4. Product evaluation

As the extruded products exited the extruder, their moisture content was accessed (MEP). After drying and cooling, the products (DEP) were packed in sealed plastic bags and stored at ambient temperature until further analysis. Their physical-chemical and technological characteristics were evaluated by their moisture (MDEP), water activity (aw), protein content (Pt), sectional expansion index (SEI), apparent density (AD), hardness (H), water absorption index (WAI), water solubility index (WSI), and CIELab system color parameters (L*, a*, b*).

2.4.1. Physical-chemical characterization

The moisture content of the blends (Mbl), the extruded products (MEP) and the dried extruded products (MDEP) was evaluated by AACC method 44-15.02 to monitor the moisture loss during the process. Water activity (aw) of milled DEPs was determined in an AquaLab 4 TE (MeterFood, Pullman-WA, USA), with milled samples. Protein content (Pt) was determined using the micro-Kjeldahl method (AACC, 2011).

2.4.2. Sectional Expansion Index (SEI) and Apparent Density (AD)

A digital caliper was used to measure extrudates' length and diameter. The measures of 20 samples were used to calculate SEI, using Eq. (4), and the AD (g/cm^3), using Eq. (5) (Ding et al., 2006).

$$SEI = d/D \tag{4}$$

$$AD = \frac{4m}{\pi d^2 L} \tag{5}$$

where d (cm) is the extrudate diameter, D (cm) is the die diameter, m is the mass (g) of an extrudate with a determined length L (cm) and diameter d (cm).

2.4.3. Instrumental Texture

The hardness (H) of the extrudates was measured using a TA-XT2i Texture Analyzer (Stable Micro Systems Ltd., Haslemere, UK) with a 50 kg load cell. The maximum force (N) required for breaking ten extrudates, measuring 5.0 cm each, was determined using a Warner Bratzler V Slot blade with a 1.0 mm/s test speed and 20.0 mm test distance.

2.4.4. Water Absorption Index (WAI) and Water Solubility Index (WSI)

WAI and WSI were determined based on Anderson (1982), with modifications. Ground extrudates (2.5 g) were suspended in 30 mL of ambient temperature water for 30 min, being vortexed for 30 s every 10 min. Afterward, the suspension was centrifuged at 3000 g for 15 min. The supernatant was transferred to aluminum Petri dishes and dried for 4 h at 105 °C. WSI corresponds to the mass of dry solids in the supernatant expressed as a percentage of the original sample weight. WAI is obtained by dividing the weight of the centrifuge residue by the dry solids of the sample.

2.4.5. Instrumental color

Instrumental color was determined by a Mini Scan XE45/0-L portable spectrophotometer (Hunter-Lab, Reston, USA), using the CIE-Lab (L*, a*, b*) system, on ground samples.

2.5. Statistical analysis

Physical-chemical and technological analysis were conducted in triplicate, unless specified. Statistical analysis was performed in the software MODDE PRO 12 (Analytics, 2017), using partial least square (PLS) regression analysis, following a PLS1 approach, to establish individual models for each response. Predictive models were based on Cox quadratic equations (Eq. (6)), considering the general mixture restriction $X_1+X_2+X_3 = 1$ and the reference mixture as the centroid of the simplex region (x'_1 1/3; x'_2 1/3, x'_3 1/3) (Eriksson et al., 1998).

$$Y = \beta_0+\beta_1X_1+\beta_2X_2+\beta_3X_3+\beta_{11}X_1^2+\beta_{22}X_2^2+\beta_{33}X_3^2+\beta_{12}X_1X_2+\beta_{13}X_1X_3+\beta_{23}X_2X_3+\varepsilon \quad (6)$$

Analysis of variance (ANOVA) was used to check for significant models and lack of fit at a 95% significance level. Models with a minimum 0.70 R^2 (goodness of fit) and a high Q^2 (goodness of prediction) were selected. The two values should not differ by more than 0.3 (Analytics, 2017; Eriksson et al., 1998). The residues' normality and the scores' distribution were analyzed for each response. The data was scaled and centered to unit variance.

3. Results and discussion

Fig. 2 shows the extruded products displayed over the sub-region of the simplex chosen for the Mixture Design. It is visible that different proportions of the components affected the extrudates' appearance and structure.

Process parameters and Blends' Moisture Content (Mbl) are presented in Table 3, while Table 4 brings the effects of blend composition on physic-chemical characteristics of extruded products and Table 5 shows effects of blend composition the technological properties of dried extruded products. Table 6 shows the significant models ($p < 0.05$) and their parameters. The correspondent contour plots are discussed under each significant response: Blends' Moisture Content (Mbl), Feed Rate (FR), Specific Mechanical Energy (SME), Extruded Products' Moisture Content (MEP), Protein Content (Pt), Sectional Expansion Index (SEI), Apparent Density (AD), and Instrumental Texture (H).

Table 3
Effects of blend composition on blends' moisture and process parameters.

Experiment	Mbl (g/100g)	FR (kg/h)	SME (kWh/kg)
1	15.46 ± 1,16	9.22	141.04
2	39.70 ± 0,64	4.35	248.97
3	17.16 ± 0,47	12.45	116.12
4	19.80 ± 0,56	10.24	127.00
5	28.15 ± 1,56	7.15	141.56
6	29.95 ± 0,36	5.81	198.94
7	19.24 ± 0,45	9.78	125.60
8	31.39 ± 0,27	6.63	141.78
9	20.68 ± 0,12	10.57	123.01
10	24.78 ± 0,47	6.77	181.48
11	23.51 ± 0,55	9.41	130.51
12	23.91 ± 0,11	8.17	132.66

Mbl: blends' moisture; FR: feed rate; SME: specific mechanical energy. Results are presented in mean ± standard deviation.

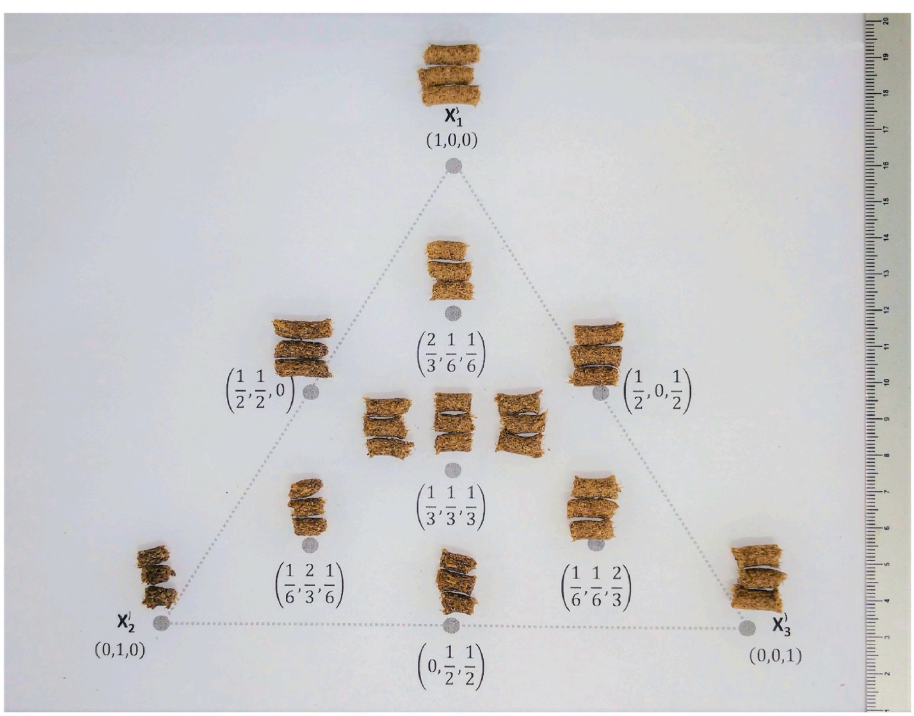

Fig. 2. Extruded products disposed over the sub-region of simplex, L-pseudocomponents x'1 = dried brewer's spent grains (BSG), x'2 = brewer's spent yeast (BSY), x'3 = corn grits (GRITS).

Table 4
Effects of blend composition on physic-chemical characteristics of extruded products.

Experiment	MEP (g/100g)	MDEP (g/100g)	Aw	Pt (g/100g)
1	9.66 ± 0,07	06.55 ± 0,27	0.5046 ± 0,01	16.71 ± 0,01
2	31.70 ± 0,51	20.45 ± 0,11	0.9004 ± 0,00	20.01 ± 0,28
3	13.02 ± 0,06	08.10 ± 0,19	0.5180 ± 0,00	13.58 ± 0,12
4	14.31 ± 0,32	11.82 ± 0,36	0.6841 ± 0,00	13.41 ± 0,2
5	22.06 ± 0,02	19.22 ± 0,82	0.8960 ± 0,00	15.53 ± 0,08
6	24.49 ± 0,18	16.12 ± 0,2	0.8419 ± 0,00	16.92 ± 0,07
7	12.44 ± 0,08	09.37 ± 0,15	0.5834 ± 0,00	16.30 ± 0,06
8	25.75 ± 0,13	17.94 ± 0,17	0.8617 ± 0,00	17.41 ± 0,28
9	14.52 ± 0,36	09.55 ± 0,51	0.5687 ± 0,01	14.74 ± 0,44
10	19.15 ± 0,21	04.93 ± 0,09	0.2761 ± 0,01	15.88 ± 0,03
11	17.83 ± 0,15	04.65 ± 0,22	0.2971 ± 0,02	15.70 ± 0,11
12	18.44 ± 0,17	05.59 ± 0,37	0.3340 ± 0,00	15.90 ± 0,23

MEP: moisture of extruded products; MDEP: moisture of dried extruded products (MDEP), aw: water activity; Pt: protein content. Results are presented in mean ± standard deviation.

3.1. *Blends: moisture content (Mbl)*

Blends with over 30 g/100 g moisture, those with the highest proportions of BSY, tended to agglomerate inside the feed hopper, at its exit, and inside the feed throat barrel. That prevented feeding the extruder at a constant rate, affecting the extrusion process and the quality of the extrudates. Feed moisture significantly affects the melt viscosity and starch modification, influencing the extrudates' physical and technological properties. Usually, increased water levels render lower melt viscosity and higher starch swelling and gelatinization, while low feed moisture is linked to higher shear rates and a higher degree of starch depolymerization and melting (Robin et al., 2011).

Mbl counter plot (Fig. 3) shows that lifting BSY levels, the highest-moisture material (86,46 g/100 g), tend to increase Mbl.

3.2. *Process parameters*

3.2.1. *Feed Rate (FR)*

The high variation in FR (Table 3) could be attributed to differences in particle size and density of the raw materials, resulting in non-homogeneous blends, and to the variation in Mbl. Both parameters were highly correlated (r = −0,92), meaning the higher the moisture content, the lower the feed rate for a fixed feed screw speed. Fig. 4 corroborates this hypothesis, showing that BSY (86,46 g/100 g moisture) had the most considerable influence over FR, leading to a drop in FR when used in high proportions. Increasing GRITS generates the

Table 5
Effects of blend composition the technological properties of dried extruded products.

Experiment	SEI	AD (g/cm^3)	H (N)	WAI (g/g)	WSI (%)	L*	a*	b*
1	1.05 ± 0,01	0.80 ± 0,04	54.00 ± 5,23	5.36 ± 0,28	2.21 ± 0,09	49.79 ± 0,47	7.62 ± 0,09	21.77 ± 0,19
2	0.94 ± 0,04	0.78 ± 0,08	97.34 ± 6,40	4.93 ± 0,24	3.30 ± 0,15	46.43 ± 1,48	7.00 ± 0,10	21.53 ± 0,32
3	1.15 ± 0,03	0.93 ± 0,04	113.58 ± 8,25	5.28 ± 0,27	2.70 ± 0,20	55.34 ± 0,75	7.59 ± 0,04	23.03 ± 0,08
4	1.10 ± 0,02	0.94 ± 0,04	138.69 ± 4,57	5.66 ± 0,52	2.38 ± 0,17	53.17 ± 0,71	8.03 ± 0,10	22.58 ± 0,52
5	1.02 ± 0,03	0.76 ± 0,05	76.72 ± 5,79	5.81 ± 0,19	2.03 ± 0,16	47.68 ± 0,43	7.27 ± 0,10	20.21 ± 0,29
6	1.02 ± 0,02	0.72 ± 0,05	84.58 ± 6,48	5.49 ± 0,19	2.86 ± 0,11	46.29 ± 0,71	7.16 ± 0,17	20.80 ± 0,51
7	1.05 ± 0,01	0.83 ± 0,04	94.02 ± 8,39	4.76 ± 0,23	2.76 ± 0,16	52.71 ± 0,55	7.74 ± 0,08	22.64 ± 0,35
8	1.03 ± 0,02	0.73 ± 0,04	100.39 ± 10,97	5.86 ± 0,09	2.97 ± 0,26	57.19 ± 14,5	7.43 ± 0,08	23.46 ± 0,24
9	1.09 ± 0,01	0.89 ± 0,03	115.03 ± 8,49	5.75 ± 0,21	2.66 ± 0,05	53.25 ± 1,08	7.63 ± 0,18	22.97 ± 0,09
10	1.01 ± 0,02	0.79 ± 0,04	92.35 ± 6,04	5.51 ± 0,15	3.22 ± 0,10	51.85 ± 1,27	7.52 ± 0,03	22.61 ± 0,10
11	1.01 ± 0,02	0.81 ± 0,03	96.08 ± 5,21	5.87 ± 0,16	3.05 ± 0,04	50.71 ± 0,43	7.59 ± 0,08	22.45 ± 0,36
12	1.05 ± 0,02	0.82 ± 0,04	96.04 ± 7,07	5.76 ± 0,05	2.80 ± 0,07	50.46 ± 0,19	7.49 ± 0,09	22.64 ± 0,08

SEI: sectional expansion index; AD: apparent density; H: hardness; WAI: water absorption index; WSI: water solubility index; L*, a*, b*: color parameters. Results are presented in mean ± standard deviation.

Table 6
Partial Least Squares (PLS) regression coefficients and model properties for significant models.

	Mbl	FR	MEP	SEI	AD	H
Constant	24.478*	0.959*	18.614*	1.042*	0.810	100.867*
DBSG	−1.478*	0.007	−1.501*	−0.001	0.120*	51.162*
BSY	5.455*	−0.079*	5.045*	−0.033*	0.050*	33.904*
GRITS	−1.173*	0.042*	0.861	0.023*	0.159	58.055*
DBSG*DBSG						
BSY*BSY					0.026*	
GRITS*GRITS						
DBSG*BSY						
DBSG*GRITS					0.029*	18.764*
BSY*GRITS						−9.389*
Response transformation		10Log (Y+1)				
DF	9	9	9	9	7	7
Q2	0.890	0.841	0.865	0.689	0.914	0.753
R2	0.969	0.898	0.946	0.844	0.969	0.930
R2adjs	0.962	0.875	0.934	0.810	0.951	0.889
RSD	1.353	0.040	1.650	0.023	0.016	6.939
p model	0.000	0.000	0.000	0.000	0.000	0.000
condition number	1.732	1.732	1.732	1.732	3.115	3.090
PLS components	1	1	1	1	4	3
p lack of fit	0.168	0.944	0.119	0.179	0.515	0.067

*Significant coefficients at a confidence level of 95%; DSG = dried spent grains; BSY = brewer's spent yeast; GRITS = corn grits; Mbl = blends' moisture; MEP = moisture of extruded products (before drying); Pt = protein content; FR = feed rate; SEI = sectional expansion index; AD = apparent density; H = hardness; DF = degrees of freedom; R^2 = coefficient of explanation; Q^2 = coefficient of prediction; R^2_{adjs} = R^2 adjusted by the degrees of freedom; RSD = residue standard deviation; p lack of fit = probability of lack of fit significant at 95% of confidence level.

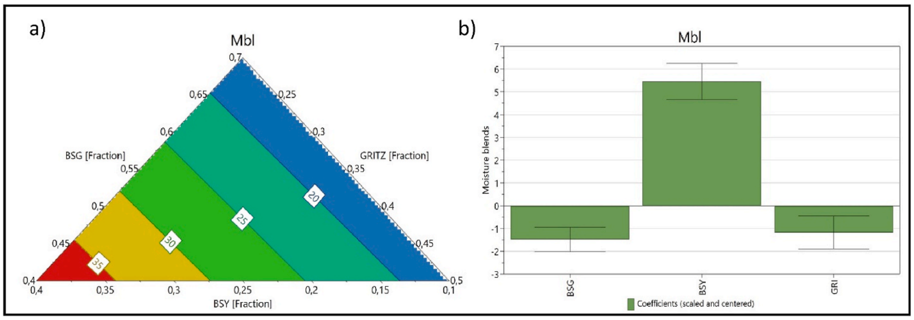

Fig. 3. Moisture of the Blends (Mbl). a) Response counter plot: cold colours (blue) indicate lower values, warmer colours (red) indicate higher values. b) PLS coefficients. BSG = dried brewer's spent grains; BSY = brewer's spent yeast; GRITS = corn grits. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

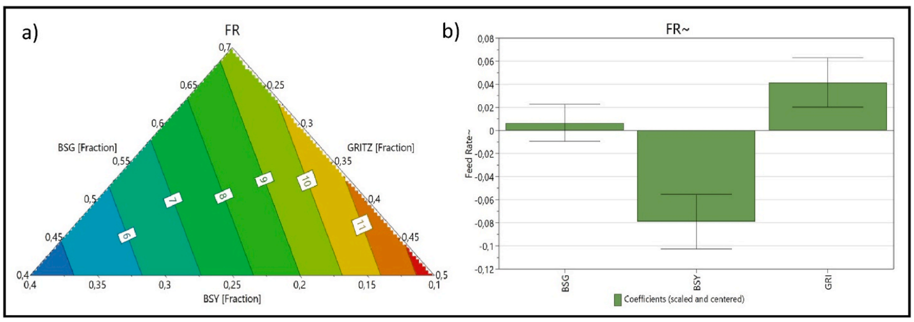

Fig. 4. Feed Rate (FR). a) Response counter plot: cold colours (blue) indicate lower values, warmer colours (red) indicate higher values. b) PLS coefficients. BSG = dried brewer's spent grains; BSY = brewer's spent yeast; GRITS = corn grits. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

opposite effect.

Notably, FR exhibited major influence over all the extrudates' technical properties, except hardness, showing correlation coefficients above 0.78 (positive or negative) with density (+), SEI (+), and MEP (−), in crescent order. Also, it was highly influenced by proteins (−), BSY (−) and Mbl (−). Therefore, it is of great importance to evaluate and control this parameter.

3.2.2. Specific Mechanic Energy (SME)

Godavarti and Karwe (1997) define SME as the amount of mechanical energy dissipated as heat per unit mass of the melt. It can be used to compare processes with different characteristics and affects the extrudates' expansion index, apparent density, and hardness. Overall, SME indicates the level of modifications – breakdown and interactions – that the material may endure during extrusion (Godavarti and Karwe, 1997; Muthukumarappan; Karunanithy, 2012; Stojceska et al., 2008).

SME depends on the screw speed (rpm), maximum motor power (kW), maximum screw speed (rpm), feed rate (kg/h) (item 2.4), and torque (%). Only the last two were dependent variables in this experiment. Torque (%), supplied by the equipment, represents the energy absorbed by the melt due to mechanical shear. It is sensitive to changes that affect the extruder flow resistance, like screw configuration and die diameter, besides the rheological properties of the melt and the feed rate (Muthukumarappan; Karunanithy, 2012; Stojceska et al., 2008).

Nascimento et al. (2017), observed a decrease in SME when higher BSG proportions and higher temperatures were employed. The authors attributed this effect mainly to the dilution of the starch material, which is responsible for higher melt viscosities due to starch gelatinization. Additionally, the high lipid content of BSG confers a plasticizer effect, lowering shear.

In this study, the blends with higher feed rates also had lower SMEs. However, PLS regression model for SME was non-satisfactory, although significant ($p < 0.05$). The model validity was 94.07 %, however, the reproducibility was low, 34.08 %, and the model triggered the skewness test.

3.3. Extruded products (MEP): moisture content

PLS regression model was significant ($p < 0,05$) and, similarly to Mbl, was impacted mainly by BSY (Fig. 5). When comparing the moisture of the products with their blends, the extrusion process reduced moisture by an average of 25.19 %, yet very heterogeneously. By analyzing the differences in moisture loss among the experiments, it was possible to associate the higher insoluble fiber content (BSG) with a higher moisture loss and the higher protein and starch contents (BSY and GRITS) with a lower loss, which corroborates the findings of Brennan (2008). In that study, the authors concluded that adding non-starch polysaccharides (NSP) would decrease the water-holding capacity of the EPs due to a faster, however weaker, binding to water associated with these components. Therefore, NSP would compete with protein and starch for the water, bind with it faster, yet release it quickly into the system.

3.4. Dried extruded products (DEP)

3.4.1. Physical-chemical characterization

The drying parameters were defined by pre-tests targeting less than 10 g/100 moisture and aw lower than 0.60 (Fellows, 2000) for the centroid of the simplex design. The moisture content of the dried extruded products (DEP) varied from 4.65 g/100 g–20.45 g/100 g, and their water activity varied from 0.2761 to 0.9004, demonstrating that the chosen conditions were not enough for all the experiments to achieve stability. The highest values were observed for exp.2 (highest in BSY) and the lowest for the centroids.

An enormous difference in moisture loss during the drying process was observed (13–74%) among the experiments. This could be due to a

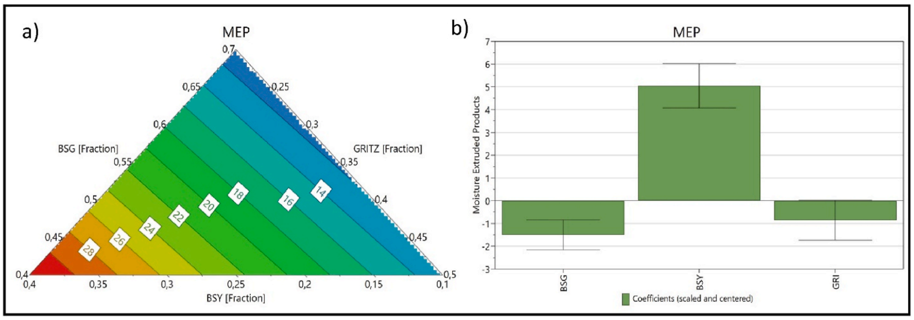

Fig. 5. MEP = Moisture of Extruded Products. a) Response counter plot: cold colours (blue) indicate lower values, warmer colours (red) indicate higher values. b) PLS coefficients. BSG = dried brewer's spent grains; BSY = brewer's spent yeast; GRITS = corn grits. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

different extent of interactions between the products' ingredients and water, as explained in the previous topic (Brennan et al., 2008). Besides, variance in heat exchange inside the oven-drier could lead to varying water loss rates.

The protein content (Pt) of DEP varied following the blends' composition. PLS regression for Pt resulted in a significant model ($p < 0.05$), however, the model was associated with a significant lack of fit and low validity.

3.4.2. *Sectional Expansion Index (SEI)*

The SEI of all the DEPs was remarkably close to 1.0, indicating low or no expansion. That could be attributed to the blends' high fiber and moisture content (Table 4). Ding et al. (2006) showed that higher feed moisture contents led to extrudates with lower expansion, higher density, and greater hardness. Robin et al. (30) demonstrated that adding insoluble fibers from various sources resulted in extruded products with reduced sectional or lateral expansion. Studies incorporating dried-BSG into extrudates prepared with different starch sources (Ainsworth et al., 2007; Makowska et al., 2013) reported similar findings.

The effect of insoluble fiber on decreasing expansion may be due to three factors: (i) steric impediment; (ii) dilution of starch; and (iii) water retention (Steel et al., 2012). It is possible to mitigate that impact by reducing feed moisture or increasing the mechanical energy (Ainsworth et al., 2007; Robin et al., 2012; Stojceska et al., 2009). Ačkar et al. (2018) suggest the use of 1 g/100 g of pectin to improve expansion in corn based expanded snacks with by-products additions.

PLS regression for SEI resulted in a significant model ($p < 0.05$). Fig. 6 shows that BSY, with high moisture, protein, and fiber content, negatively influenced SEI, while GRITS, high in starch - the main structural component associated with extrudates expansion, showed a positive impact.

3.4.3. *Apparent Density (AD)*

Higher small-cell density is expected for extrudates with the addition of insoluble fibers, although the type of fiber and the process conditions also influence product bulk density (Robin et al., 2012). Ainsworth et al. (2007) observed an increase in AD from 0.20 to 0.89 g/cm^3 when adding 30 g/100 g BSG, using a screw speed of 100 rpm. At 300 rpm, however, AD ranged from 0.12 g/cm^3 (0 g/100 g BSG) to 0.35 g/cm^3 (30 g/100 g BSG), highlighting the importance of optimization of process parameters for achieving good quality high-fiber extrudates. In our study, the density of the products ranged from 0.72 g/cm^3 (Exp. 6) to 0.94 g/cm^3 (Exp. 4).

The high fiber content interferes with the viscoelastic properties of the melt, breaking the structure of the continuous starch phase and increasing its apparent viscosity in the section before the die. That may cause increased resistance to the growth of bubbles at the exit of the equipment, provoking a decrease in the expansion index and an increase in the apparent density. Also, the melt elasticity decreases with increased fiber content, probably due to the low adhesion properties at the interface between insoluble fibers and starch, caused by their low chemical compatibility (Robin et al., 2012).

All these factors, alongside steric interference from the fiber particles, contribute to an increment in rupture points in the bubble membranes, leading to their burst when exiting the extruder. Therefore, extrudates with high insoluble fiber content often manifest a porous surface (Robin et al., 2012).

The PLS regression for density resulted in a significant model ($p < 0.05$). Unexpectedly, the correlation between AD and SEI was positive – so extrudates with a lower SEI also showed lower AD. Besides, lower AD values were achieved for higher BSY and BSG content, as illustrated in Fig. 7.

A possible explanation for that could be due to the positive correlation between apparent density (AD) and feed rate (FR): at the same feed screw speed, blends with higher moisture, which also means higher

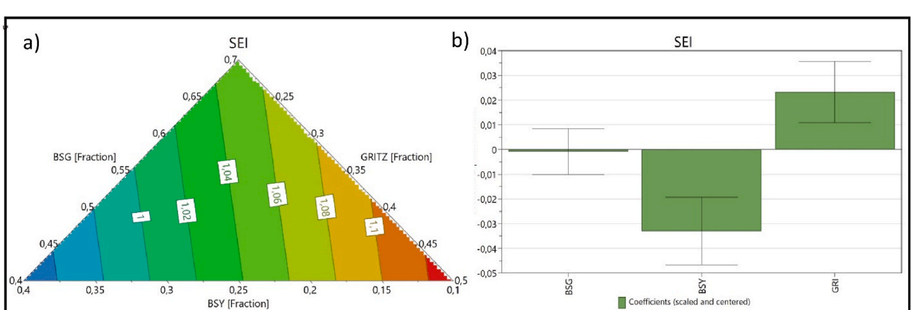

Fig. 6. Sectional Expansion Index (SEI). a) Response counter plot: cold colours (blue) indicate lower values, warmer colours (red) indicate higher values. b) PLS coefficients. BSG = dried brewer's spent grains; BSY = brewer's spent yeast; GRITS = corn grits. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

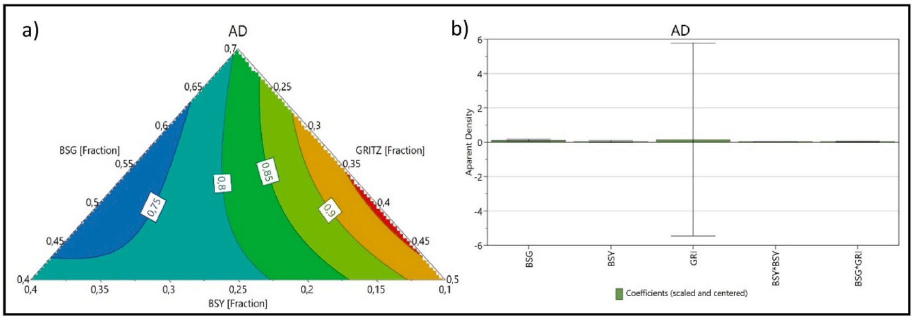

Fig. 7. Apparent Density (AD). a) Response counter plot: cold colours (blue) indicate lower values, warmer colours (red) indicate higher values. b) PLS coefficients. BSG = dried brewer's spent grains; BSY = brewer's spent yeast; GRITS = corn grits. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

BSY percentage, achieved lower FR, and, consequently, lower outflow rate, resulting in a product with lower mass by volume unit (density).

3.4.4. Instrumental Texture

The extruded products exhibited high hardness values, 54.00–138.69 N, compared to other studies that used lower BSG contents. Ainsworth et al. (2007) achieved values from 11.5 to 14.44 N (0–30 g/100 g BSG, 300 rpm), while Stojceska et al. (2008) found the highest hardness values for the highest BSG additions (30 g/100 g BSG): 23.3 N (350 rpm) and 33.2N (150 rpm). Ačkar et al. (2018), using a laboratory single-screw extruder, decreased hardness by nearly 75 % by adding 1 g/100 g pectin to the formulations with 30 g/100 g BSG.

According to Robin et al. (2012), extruded products' mechanical properties depend on the continuous phase's characteristics - associated with the physic-chemical properties and cell-wall thickness - and the dispersed phase characteristics - porosity, density, and cell distribution. Adding insoluble fiber generally leads to an increase in small cells with thicker cell-walls, resulting in higher bulk density and forming structures with higher resistance, consequently, higher hardness.

In our results, hardness positively correlated with density (0.66). Nonetheless, as shown in the contour plot, Fig. 8, the lowest hardness values were achieved by the maximum BSG additions, possibly due to the fragile and brittle structure conferred by an ingredient high in fiber and low in starch.

The dilution of starch and the disturbance in the continuous phase by the large fiber particles could be responsible for this, rendering low cohesiveness. Although most authors believe high hardness values are not appreciated, Nascimento et al. (2017) consider that the high fiber content appeal could counterbalance that.

3.4.5. Water Absorption Index (WAI) and Water Solubility Index (WSI)

WAI and WSI methods were developed to evaluate starch functionality after extrusion. WAI represents the water absorbed by the sample, mainly by the gelatinized starch, while WSI stands for the percentage of molecules soluble in water and is associated with process severity and the degree of starch degradation. Both parameters showed scattered values with no significant model, possibly due to the high fibre and low starch levels (Ainsworth et al., 2007; Colonna et al., 1989; Robin et al., 2011).

WAI presented similar values to the ones obtained by other studies with the addition of BSG or other fibres (Ainsworth et al., 2007; Stojceska et al., 2008, 2009). According to Robin et al. (2011), insoluble fiber binds to water, restricting water for starch gelatinization and, therefore, decreasing WAI.

Adding BSG could result in diminished pressure and mechanical intensity of the process due to its high amounts of lipid and insoluble fiber, besides low starch content (Makowska et al., 2013). Therefore, the high levels of BSG plus the high moisture of the blends may be responsible for the low WSI (2.03–3.30 %) since they reduce mechanical shear and melt viscosity, resulting in low starch depolymerization (Makowska et al., 2013; Stojceska et al., 2009; Robin et al., 2011).

3.4.6. Instrumental color

The instrumental color of the extruded products showed slight variation (Table 5), indicating that varying the proportions of BSG, BSY and GRITS, in the levels applied in this study, did not affect color. Other studies showed a decrease in lightness (L*) with increasing proportions of the BSG (Ainsworth et al., 2007; Stojceska et al., 2008). Adding insoluble fiber usually results in products with smaller mean cell areas, lower SEI, and higher AD, which are associated with darker products. Moreover, higher moisture content and barrel temperatures usually lead to lower lightness (Oliveira et al., 2017).

4. Conclusions

In this exploratory study, we opted to dry BSG before the

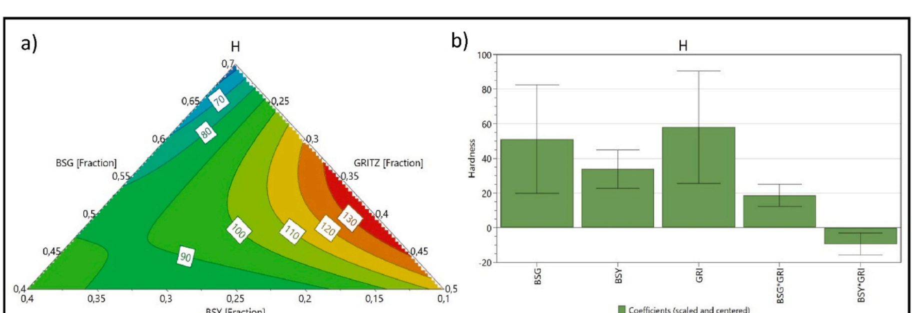

Fig. 8. Hardness (H). a) Response counter plot: cold colours (blue) indicate lower values, warmer colours (red) indicate higher values. b) PLS coefficients. BSG = dried brewer's spent grains; BSY = brewer's spent yeast; GRITS = corn grits. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

thermoplastic extrusion to maximize by-product incorporation. Therefore, we could design blends containing 40–70 g/100 g BSG, 10–30 g/100 g BSY and 20–40 g/100 g GRITS, with up to 30 g/100 g moisture. The extruded products presented high protein concentration (13–20 g/100 g), low expansion (0.94–1.15), and high density (0.73–0.94 g/cm^3), parallel to a high variation in the values of hardness (54–138 N) and moisture content (4.65–20.45 g/100 g). Overall, thermoplastic extrusion showed feasibility for repurposing large volumes of brewer's by-products and reducing waste.

From the results of this work, it is possible to choose commercial, nutritional, and technological targets to develop an optimized product to meet consumer needs for sustainable and healthier products. The extrudates could be made into flour and added to granolas, cereal bars, snacks, or other innovative products. However, before use, studies must include microbiological, toxicological and sensory analyses. Nutrient biodigestibility and bioavailability, and nucleic acid content should also be determined for nutritional purposes. And assessing paste and melt viscosity of blends and products may help to have a better understanding of the effects of the extrusion process.

CRediT authorship contribution statement

Aline D.C. Brito: Conceptualization, Investigation, Methodology, Data, Formal analysis, Data interpretation, Software, Supervision, Writing – original draft, Writing – review & editing. **Julie S. Jisaka:** Investigation. **Aline C.R. Pereira:** Investigation. **Caroline J. Steel:** Conceptualization, Writing – review, Supervision, Project administration.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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